in a review of current research in what is known as "isostasy." In the discussion of this puzzling question, Mr. Willis advances the theory that the foundation of all the continents is composed of solid rock which is self-crushed to a depth of about 120 kilometers, but rendered sufficiently rigid by pressure to maintain its form during prolonged geologic periods with but slight change. In spite of stresses occasioned by erosion of continental reliefs, this mass is capable of movements sometimes resulting in the gradual elevation of continents and the more vigorous uplifting of mountains, through which isostatic equilibrium is restored.

In line with the construction and condition of the globe, another author, Professor Thomas Chrowder Chamberlin, brings up the further vital question, "The Future Habitability of the Earth," in an article in which he reviews the past, and considers the future, of the world as a dwelling place for the human race. Many branches of science enter into the discussion, but upon geology, physics, chemistry, astronomy and astrophysics rests the burden of the arguments. Mr. Chamberlin thinks that the earth will remain habitable for tens of millions of years, but concedes that the close approach of a celestial body to the sun would probably result in the disruption of the solar system and bring disaster to the earth. He further states, in regard to the future possibilities of scientific research, that "when moral purpose and research come to be the preeminent characteristics of our race by voluntary adoption and by the selective action of the survival of the fittest, and when these most potent attributes join in an unflagging endeavor to compass the highest development and the greatest perpetuity of the race, the true era of humanity will really have been begun."

Several papers come under the head of botany, among them an interesting sketch of the sacred ear-flower of the Aztecs, a plant whose identity has been a mystery for years and only recently rediscovered by the author, Mr. W. E. Safford, of the Bureau of Plant Industry. This little flower, resembling the human ear, has a remarkable history and dates back to the early explorations of Mexico. It was first described in 1569, by Padre Bernardino de Sahagun, who states that it was much used owing to its delicious fragrance and its flavor when used as a spice. Despite the formidable name, Xochinacaztli, which it bears, the author suggests its cultivation on account of its unusual fragrance and pleasant spicy flavor. Mr. Henry S. Graves, chief of the Forest Service, contributes a well-illustrated and original article on forest preservation, in which he carefully considers all points in the great problem, making many things clear which have long been obscure.

Those interested in medical research and allied subjects will find matter of concern in the following papers: "Manifested Life of Tissues Outside of the Organism," by Alexis Carrel and Montrose T. Burrows; "Epidemiology of Tuberculosis," by Robert Koch; "The Significance of the Pulse Rate in Vertebrate Animals," by Florence Buchanan, D.Sc., and "Sanitation on Farms," by Allen W. Freeman, M.D.

A comprehensive paper on the contemporary Slav peoples, from a geographical and statistical point of view, by Ludor Niederle, of the Bohemian University of Prague, which has been translated from the Slavic language into English, furnishes new information on the history and distribution of these peoples. Dr. J. Walter Fewkes, of the Bureau of American Ethnology, contributes a brief review of his recent work and investigations in cave dwellings, both at home and abroad. This paper is entitled "The Cave Dwellings of the Old and New World."

The Report also contains biographies of Melville Weston Fuller, Sir Wm. Huggins and Alexander Agassiz, together with papers on several other subjects treated by competent authors, many of whom are world-wide authorities.

SPECIAL ARTICLES

CESTODE CELLS IN VITRO

THE desirability of throwing any light whatsoever upon the question of the character of

cell-division in cestode cells prompted me to attempt to apply the method of Harrison[1] in growing neurones, to cultures of cells from the tape-worms which are available from the spiral valve of the dog-fish, sand-shark, the cystic duct of the squeteague, etc. Inasmuch as sand-sharks were obtained only infrequently during the summer of 1911 at the Marine Biological Laboratory, Woods Hole, Mass., I was forced to depend upon the small *Calliobothrium* of the dog-fish, rather than upon the larger and more desirable *Crossobothrium* of the sand-shark. The form in the squeteague was not obtained in sufficient numbers to be of much use in the experiments.

I was directly led to the attempt of growing the isolated cells of the tape-worm from an examination which Dr. Frederic M. Hanes and Dr. R. A. Lambert kindly permitted me to make, of a series of their preparations of mouse sarcoma cells growing *in vitro* at the College of Physicians and Surgeons, New York. The marked success which they experienced[2] along this line, where the cells grew out from the small pieces of tissue from the living mouse and exhibited amœboid locomotion, absorption of granules of carmin, presence of mitosis, etc., seemed possible in other material.

The method which was used in the present set of experiments was as follows: Slides with depressions and their covers were sterilized in a hot-air oven at 200° C. for ten minutes. Where the plasma of the blood of the dog-fish or sand-shark was used, it was centrifuged after its drainage from the caudal artery under aseptic conditions (canula sterilized in olive oil at boiling) in paraffin lined tubes surrounded with freezing mixture and the supernatant plasma was pipetted into cooled tubes which were kept in an ice box until used. The precautions taken to insure sterility were of course aimed at keeping the plasma as free from bacterial decomposition as possible and thus offer to the cells of the tape-worm as nearly isotonic a medium for growth as practicable. If *Crossobothrium* was used as material, the blood of the sand-shark was used; if *Calliobothrium*, the dog-fish blood.

The tape-worms were invariably taken from a fish which had just been killed. They were washed off in sterile sea-water, with several changes and finally teased into small bits of groups of cells which were then transferred to a drop of serum laid upon a cover-glass with a platinum loop. The cover was then mounted in vaseline over the depression. In case *intra vitam* stains were used, these were introduced at once before the preparation was sealed. Inasmuch as I was dealing with a poikilothermous animal and not with a constant temperature, warm-blodded one, as in the work of Harrison, Burrows,[3] Lambert and Hanes *et al.*, it was not necessary to keep the slides in a thermostat. The temperature, however, varied only from about 17° C. to 23° C.

The cells prepared in this manner began to migrate from the mass within twelve hours and were to be found at the end of that period, or earlier, in some cases, distributed over the whole of the culture medium. I could not observe any true amœboid motion, as evidenced by the formation of pseudopodia and the streaming of granules, but there could be no other factor operating except the locomotion of the cells as far as I could determine. It must be understood that the cells are very small, and exhibit a marked degree of refraction, as is the case with all of the cestodes and even if blunt pseudopodia were formed, they would be observed only under the most favorable conditions, while probably protoplasmic streaming would be impossible to see. Moreover, when the plasma of *Limulus* was used, this distinction of cells was not observed, owing probably to the toxic action of the copper content of this peculiar blood.

Saline solutions of various content and pure sea-water were also used as media and in

[1] Harrison, R. G., 1910, *Journ. Exper. Zool.*, 9: 787.

[2] Lambert and Hanes, 1911, *Journ. Am. Med. Assoc.*, three communications.

[3] Burrows, M. T., 1910, *Journ. Am. Med. Assoc.*, p. 2057.

the latter, excellent results were obtained[4] as far as the living of the cells is concerned, but the unfortunate condition arose invariably that a large spirochæte *Spirochæte balbanii,* perhaps. grew in such numbers that the cells were soon covered with their growths and disintegration ensued. All attempts at freeing the medium from this animal, such as the use of $HgCl_2$ in various dilutions with the cestode before the tissue was teased were of no avail and the experiments in this direction were abandoned.

Difficulty was experienced in regard to the invasion of bacteria and spirochætes into the plasma mounts, but their growth for some reason was not as pronounced as with seawater and indeed in many cases, no bacteria or spirochætes were found after ten days time. Therefore the plasma method was used throughout the experiments.

It was expected that the cells, which could be made to live in apparently good condition for several weeks in the plasma medium would undergo the process of fission and during the earlier stages of the study, I was convinced that I was observing such a phenomenon, but I finally was driven to the conclusion that what I saw was in no case cell division, but rather an association of the cells in twos which resembled a cell undergoing reproduction. I am under the impression that the cells which are cultured in this way do not undergo cell-division at all. I made charts of the slides which I had under observation for a week at a time and checked the behavior of all of the cells in each slide with camera lucida drawings, comparing those made one day with those made on the day previous. If any increase in number of the cells had occurred, I should have noticed it, of course.

In order that the cells may be stained for nuclear contents, it was necessary to take down the preparation and float the cover upon a saline solution for an hour so that the fibrin should dialyze out and I am under obligation to Professor Harrison for this technique, which is quite necessary, for otherwise the fibrin takes the nuclear stain to such a degree that the cells of the cestode become indistinguishable. Heidenhain's iron-hematoxlin, bismarck brown (used as an intra-vitam stain, as well as for fixed preparations), Ehrlich's hematoxylin, safranin and other stains were used, but all of them gave consistent results to show that cell-division was not taking place. The preparations were in some cases dried in the air or over a flame and mounted directly after staining. In other cases, the covers were inverted over osmic fumes and then passed through the alcohols into balsam.

Typical resting nuclei were seen in all of the preparations, but no indication of cell-division could be determined. It may be that cell-division occurs in rhythms as indeed is the case in *Crossobothrium* according to the results of W. C. Curtis and it may be that the period over which these present experiments extended did not cover the fission period. They were begun July 5 and extended to August 15.

In regard to the matter of regions in the cestode where fission is more likely to occur, such as the region immediately posterior to the scolex, in the "growth regions" of *Crossobothrium,* described by Curtis and in the maturing sex products, it may be said that all of these regions were examined by the methods described in this communication. No difference could be determined between the cells from one region and those from another.

While the results were unsatisfactory as far as the end desired is concerned and while they are negative throughout, it seems to me that the method may be applied to more favorable material where the character of cell-division is in question. The unmistakable presence of mitosis in the sarcoma tissue investigated by Lambert and Hanes, where amitosis has been so frequently described, is an instance in point. The "amitosis" of the follicle cells of insects is desirable and ever available material for such an investigation.

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Hartford, Conn.,
November 1, 1911

[4] Compare Lewis and Lewis, 1911, *Anatomical Record,* 5: 277.